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EFFECT OF HUMIC ACID AND WATER QUALITY ON PEROXIDASE AND CATALASE ENZYMES ACTIVITY IN LEAVES OF DATE PALMS CV. BARHEE

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ABSTRACT
The present research was conducted during the years of 2013 and 2014. Twenty four young date palm trees of three years old were selected for Barhee cv. which propagated by tissue culture and that cultivated in the orchard of tissue culture date palm station / Ministry of Agriculture that is located in the desert region of Alnajaf Alashraf Governorate/ Iraq. The aim was to study the effect of organic humic acids on Peroxidase and Catalase Enzymes activity in leaves of date palms. Four treatments of Humic acid 80 % : 0, 2, 4 or 6 g. L^{-1} and their solutions were used by the addition to the soil around the trees of monthly intervals for sixteen months started on 6 / 2 / 2013 with the use of two irrigation water quality (well water or river water). The measurement on 1 /6 / 2014 indicated that the addition of humic acid or well water treatments caused significant increases in Peroxidase and Catalase activity. Treatments of using humic acid with well water irrigation also caused increases in Peroxidase and Catalase activity, on the other hand the river water showed the reverse values for well water.

KEY WORDS: Date palm, Humic organic acids, Salt stress, Desert region cultivation, enzymes.

INTRODUCTION
Date palm (*Phoenix dactylifera* L.) is one of the important trees grown in Iraq. Previously, Iraq was ranked the first in the cultivation and product of date palms, but because of many factors, including high salinity in the soil and / or irrigated water decreased the number of palm trees (Al-Najar, 2008). According to Tester and Davenport (2003); Flowers (2004) and Al-Aamiry (2014) increasing of salinity levels in the soil or irrigated water caused decreases in vegetative growth of plants. Kassem, 2012 and EL-Khawaga (2013) found that the addition of humic organic acids caused a significant increases for vegetative growth of plants. However, it is minimizes the salinity stress and led to increased the tolerance by its role on improve the soil condition and roots absorption witch effected on some physiological properties in the leaves. This experiment aims to investigate the effect of humic organic acids on Peroxidase and Catalase enzymes activity in pinnae of date palms that planted in the desert regions which irrigated with well water that displays the trees to salinity stress .

MATERIALS & METHODS
A field experiment was conducted in an orchard belong to the tissue culture date palm station/ Ministry of Agriculture that located in the desert region of Al-Najaf Al-Ashraf Governorate/ Iraq at the years of 2013 and 2014. The experiment was done on twenty four young date palms (*Phoenix dactylifera* L.) trees of three years old of Barhee cultivar that propagated with tissue culture and planted in a straight lines with a dimension of 5 × 5 meters. Four treatments with different concentrations of humic acid 80 %: 0, 2, 4 or 6 g.L^{-1} (diluted with one letter of distilled water) by the addition of its solution to the soil around the trees at monthly intervals for sixteen months started on 6/ 2/ 2013 with using two irrigation water quality (well water or Euphrates river water) by dripping system. Weeding practices were done equally when it considered necessary. The chemical and physical characteristics for the orchard soil and chemical analysis for water quality and humic acid are presented in table (1).

TABLE 1: Chemical and physical properties of soil, chemical analysis for well, river water and humic acid before treatment application

Characters		Soil	Well water	River Water	Humic acid
pH		7.41	7.11	7.96	7.08
EC		5.64 $dS.m^{-1}$	6.44$dS.m^{-1}$	1.24$dS.m^{-1}$	---------
CEC		4.70 c. $mol.kg^{-1}$	---------	---------	---------
Organic matter		0.379 %	---------	---------	---------
Available macronutrients	N	4.16 $mg.kg^{-1}$	0.98 $mg.L^{-1}$	0.61 $mg.L^{-1}$	4.87 %
	P	6.50 $mg.kg^{-1}$	3.82 $mg.L^{-1}$	2.11 $mg.L^{-1}$	0.01 %
	K	11.16 $mg.kg^{-1}$	6.62 $mg.L^{-1}$	0.21 $mg.L^{-1}$	11.21 %
Soluble ions	Ca	18.84 $meq.L^{-1}$	8.52 $meq.L^{-1}$	1.64 $meq.L^{-1}$	0.50 %
	Mg	17.20 $meq.L^{-1}$	10.81 $meq.L^{-1}$	2.85 $meq.L^{-1}$	0.22 %
	Na	37.23 $meq.L^{-1}$	44.14 $meq.L^{-1}$	3.72 $meq.L^{-1}$	---------
	HCO3	5.84 $meq.L^{-1}$	7.56 $meq.L^{-1}$	1.23 $meq.L^{-1}$	---------
	SO4	13.41 $meq.L^{-1}$	8.90 $meq.L^{-1}$	1.56 $meq.L^{-1}$	---------
	Cl	34.33 $meq.L^{-1}$	46.11 $meq.L^{-1}$	6.20 $meq.L^{-1}$	---------
Sodium adsorption ratio (SAR)		8.77	14.20	2.53	---------
Particle size distribution	Sand	914.65 $g.kg^{-1}$			
	Silt	72.11 $g.kg^{-1}$			
	Clay	13.24 $g.kg^{-1}$	---------------------------------		
	Texture	Sandy			

Studied Parameters: Determination of the effects of enzymes was done on 1 /6 / 2014 on leaves included the following:

1. Activity of Peroxidase: Was determined according to the Mustafa (2003) by using spectrophotometer at optical absorption of 470 nm. and calculated according the equation

Peroxidase activity ($unit.g^{-1}$) =Absorption/ (W / Reaction volume) × V

2. Activity of Catalase: Was determined according to the Aebi (1983) by using UV. Spectrophotometer at optical absorption of 240 nm. and calculated according the equation : (Frary *et al*., 2010)

Catalase activity ($unit.g^{-1}$) = (Δ Absorption/ mni.) × Reaction volume / 0.001

The experiment design was factorial with two factors adopted with Randomized Complete Block Design (RCBD). Least Significant Differences Test was used to compare means when it considered significant at probability of 0.05 (Al-Rawi and Khalaf-Allah, 2000).

RESULTS

1. Activity of Peroxidase**:** Results in table (2) showed that there was a significant difference between the concentrations of humic acids, the treatment of (6 g. L^{-1}) gave the highest value for Peroxidase activity in the leaves of 303.8 $units.g^{-1}$ compared with control treatment that gave 262.5 $units.g^{-1}$. Moreover, water quality significantly affected Peroxidase activity and gave the highest value of 301.3 $unit.g^{-1}$ with well water. The interactions among both factors (humic acids and water quality) showed a significant variance in Peroxidase activity, and concentration of humic acids 6 g. L^{-1} with well water gave the highest Peroxidase activity of 316.65 $unit.g^{-1}$ while, plants of control treatment irrigated with river water gained the lowest values of Peroxidase activity of 251.0 $unit.g^{-1}$.

2. Activity of Catalase: Table (3) showed a significant difference between concentrations of humic acids ,the differences were found clearly in the treatment of (6 g. L^{-1}) that gave the largest values for Catalase activity that gave 70.53 $unit.g^{-1}$ compared with plants of control treatment that gave 39.48 $unit.g^{-1}$. On the other hand, water quality showed a significant effect on Catalase activity. Well water gave the highest values of 64.94 $unit.g^{-1}$, while, river water gave 57.73 $unit.g^{-1}$. The interactions among both factors showed significant differences in Catalase activity. The concentration of humic acids 6 g. L^{-1} with well water gave the highest Catalase activity of 75.13 $unit.g^{-1}$, whereas, plants of control treatment irrigated with river water which revealed the lowest values of Catalase activity of 39.10 $unit.g^{-1}$.

DISCUSSION

Data in tables (2 and 3) showed that, Peroxidase and Catalase activity ($unit.g^{-1}$) in leaves were significantly higher in all concentrations of the humic acids; it increased with the increase in the concentrations. The reason for this increase could be due to the rolls of humic acids to increase in the physiological and biochemical processes at the cell sap and the formation of protein which lead to an increase in the concentrations and activation of the enzymes at the cells (Dantas *et al.*, 2007), in addition to its effects in increasing the ability of roots absorptions of water and nutrients ions that help increases cations exchange capacity (CEC), the formation of natural chelating compounds and facilitation nutrients transport process , especially potassium with the important role in the activation of protein and enzymes Levitt (1980); Taiz and Zeiger (2006) and Abada *et al.* (2010). It can be seen that activity of the enzymes was increased significantly by using well water in irrigation , this increase can be attributed mainly to the ability of the plant to resist salinity and oxidative stress, especially damage resulting from certain types of active oxygen species ($O2^-$, H_2O_2, OH)by increase production of antioxidants enzymes (Peroxidase and Catalase) in the leaves tissues which have a valuable role to minimize these types of active oxygen Dat *et al.* (2000) ; Mittler (2002) and Munns and Tester (2008). These results were in agreement with Al-Aamiry (2014) in date palms leaves and with Hasan (2005) in olives leaves .

TABLE 2: Effect of Humic Acid, Water quality and Their Interactions on Peroxidase activity in the leaves of Date palms cv. Barhee

Treatments			Activity (unit. g
	(0) Control		262.5d
	2 g. L(		272.5c
	4 g. L(		292.5b
Humic Acid	6 g. L(		303.8a
Quality of water	Well water		301.3a
	River water		277.2b
	0)(	Well water	274.0e
		River water	251.0g
	2 g. L(	Well water	283.0d
Humic Acid		River water	262.0f
×	4 g. L(	Well water	304.0b
Quality of water		River water	281.0d
	6 g. L(	Well water	316.6a
		River water	291.0c

TABLE 3: Effect of Humic Acid , Water quality and Their Interactions on Catalase activity in the leaves of Date palms cv. Barhee

Treatments			Activity (unit. g
	(0) Control		39.48c
	2 g. L(		64.40b
	4 g. L(		67.08ab
Humic Acid	6 g. L(		70.53a
	Well water		64.94a
Quality of water	River water		57.73b
		Well water	39.87e
	0)(	River water	39.10e
	2 g. L(	Well water	71.30ab
		River water	57.50d
	4 g. L(	Well water	72.83a
		River water	61.33cd
Humic Acid	6 g. L(	Well water	75.13a
×		River water	65.93b
Quality of water			

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